



APPLICANT(S): Fenical et al.
TITLE: SALINOSPORAMIDES AND METHODS FOR USE
THEREOF
Application No.: 10/600,854 File Date: June 20, 2003
Docket No.: UCSD1530-2

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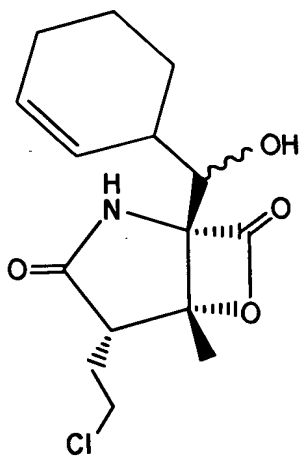


FIG. 1

Phylogeny of MAR1

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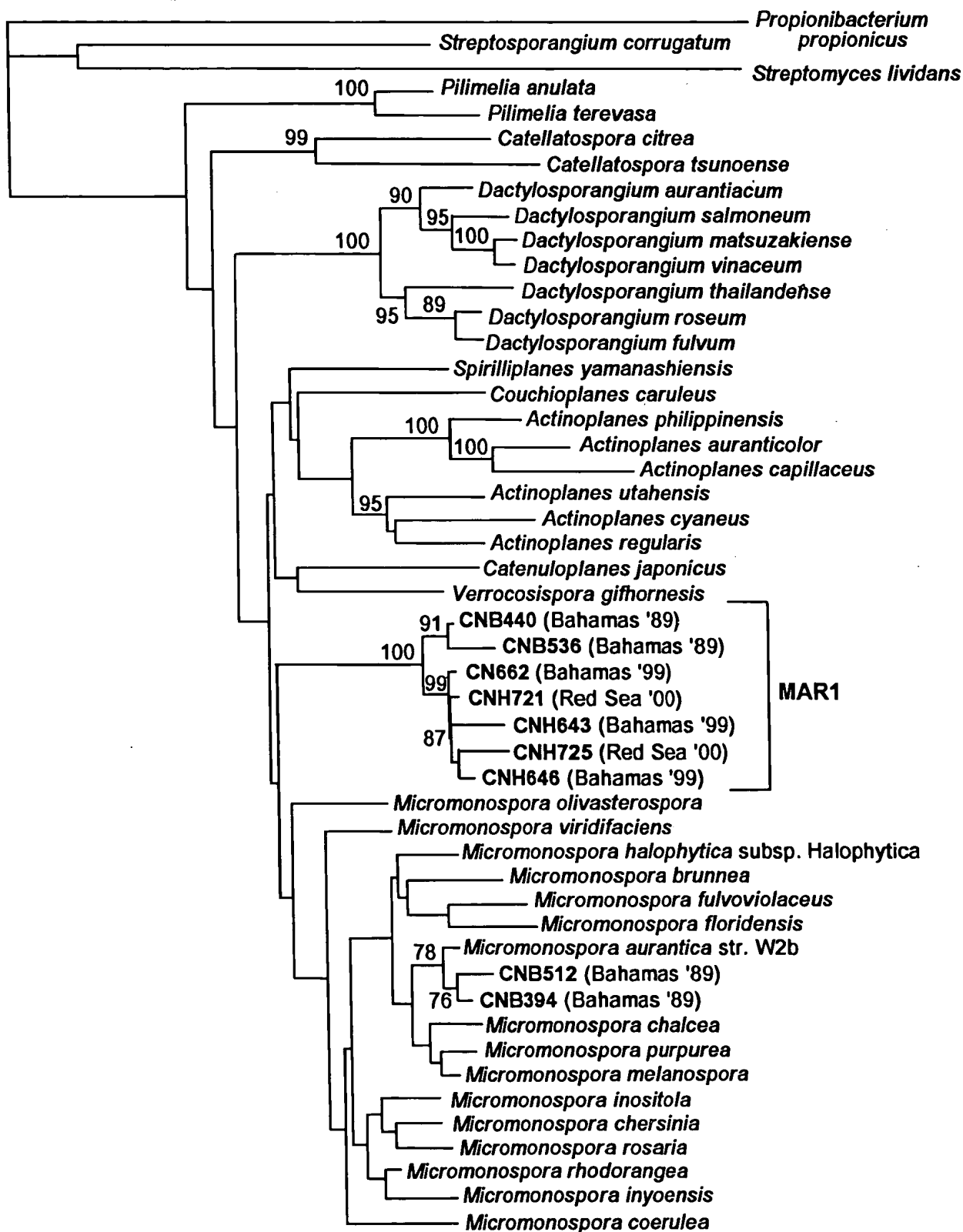


FIG. 2

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Etoposide (2)

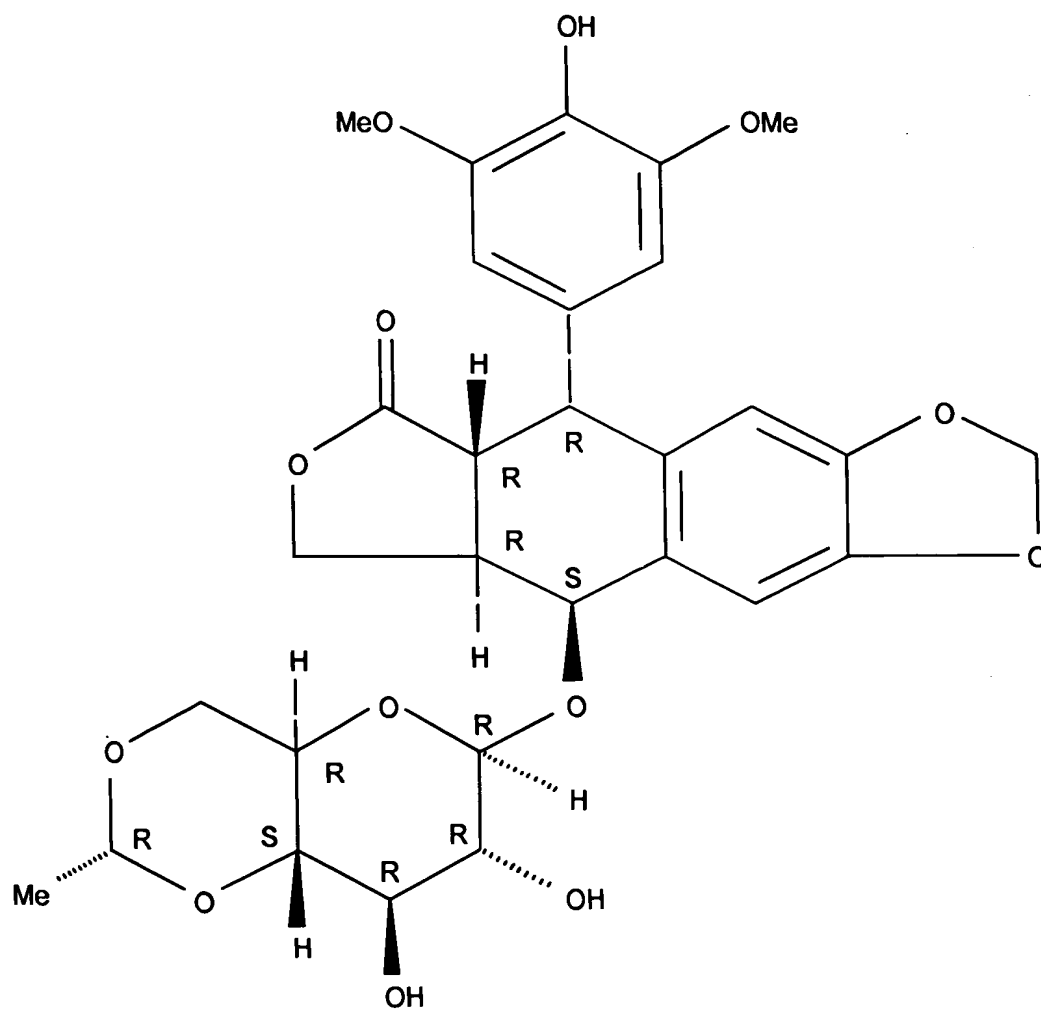


FIG. 3

Cytotoxic activity of salinosporamide A (1) and dose response curve 4/15

Comp8 (units/ml)

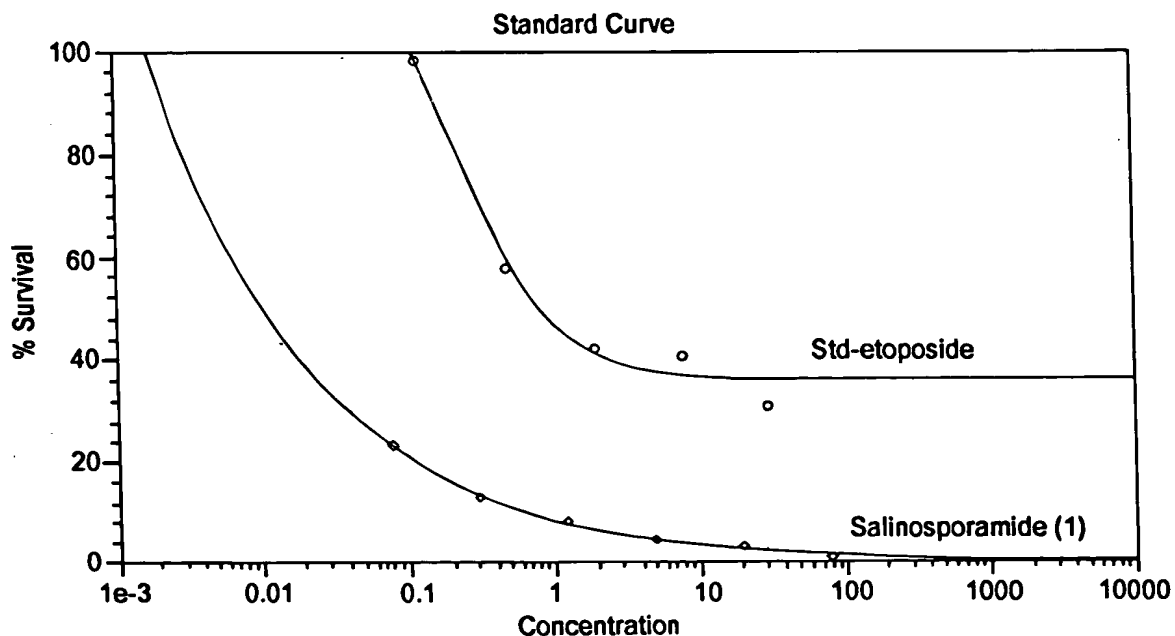
Sample	Wells	Sample #	OD	Mean Value	Ctr. Avg OD	% Survival	Concentration	IC50
Co03	C8	1	0.013	0.013	1.305	0.958	78.125	0.011
Co04	D8	2	0.044	0.044	1.305	3.335	19.531	0.011
Co05	E8	3	0.059	0.059	1.305	4.484	4.883	0.011
Co06	F8	4	0.105	0.105	1.305	8.011	1.221	0.011
Co07	G8	5	0.170	0.170	1.305	12.993	0.305	0.011
Co08	H8	6	0.304	0.304	1.305	23.266	0.076	0.011

Standards (mg/ml)

Sample	Concentration	Back Calc Conc	Wells	OD	Mean OD	% Survival	CV %	Ctr. Avg OD	IC50
Sta03	31.250	Range?	C12	0.413	0.413	31.621	0.0	1.305	0.828
Sta04	7.813	Range?	D12	0.542	0.542	41.510	0.0	1.305	0.828
Sta05	1.953	Range?	E12	0.557	0.557	42.660	0.0	1.305	0.828
Sta06	0.488	Range?	F12	0.760	0.760	58.222	0.0	1.305	0.828
Sta07	0.122	Range?	G12	1.287	1.287	98.620	0.0	1.305	0.828
Sta08	0.031	Range?	H12	1.615	1.615	123.764	0.0	1.305	0.828

Smallest standard value: 0.413

Largest standard value: 1.615



	A	B	C	D	R ²
◊ Comp.8 (Comp8: Concentration vs % Survival)	2183.255	0.398	8.34e-7	0.073	0.997
◊ Std (Standards: Concentration vs % Survival)	133.236	1.227	0.192	36.119	0.993

FIG. 4

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Isolation of salinosporamide A (1), separation scheme

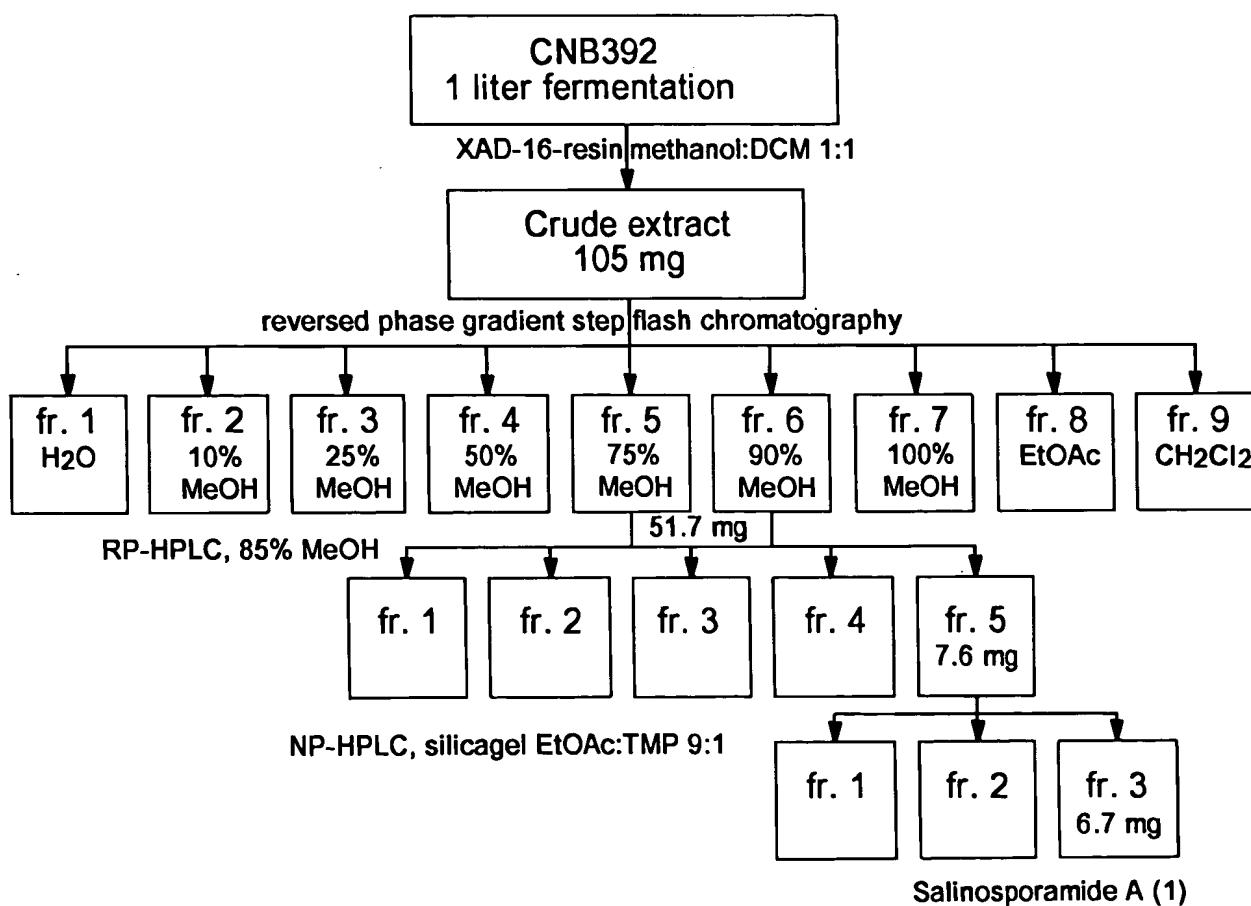


FIG. 5

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¹H-NMR of salinosporamide A (1), 300 MHz, d5-pyridine

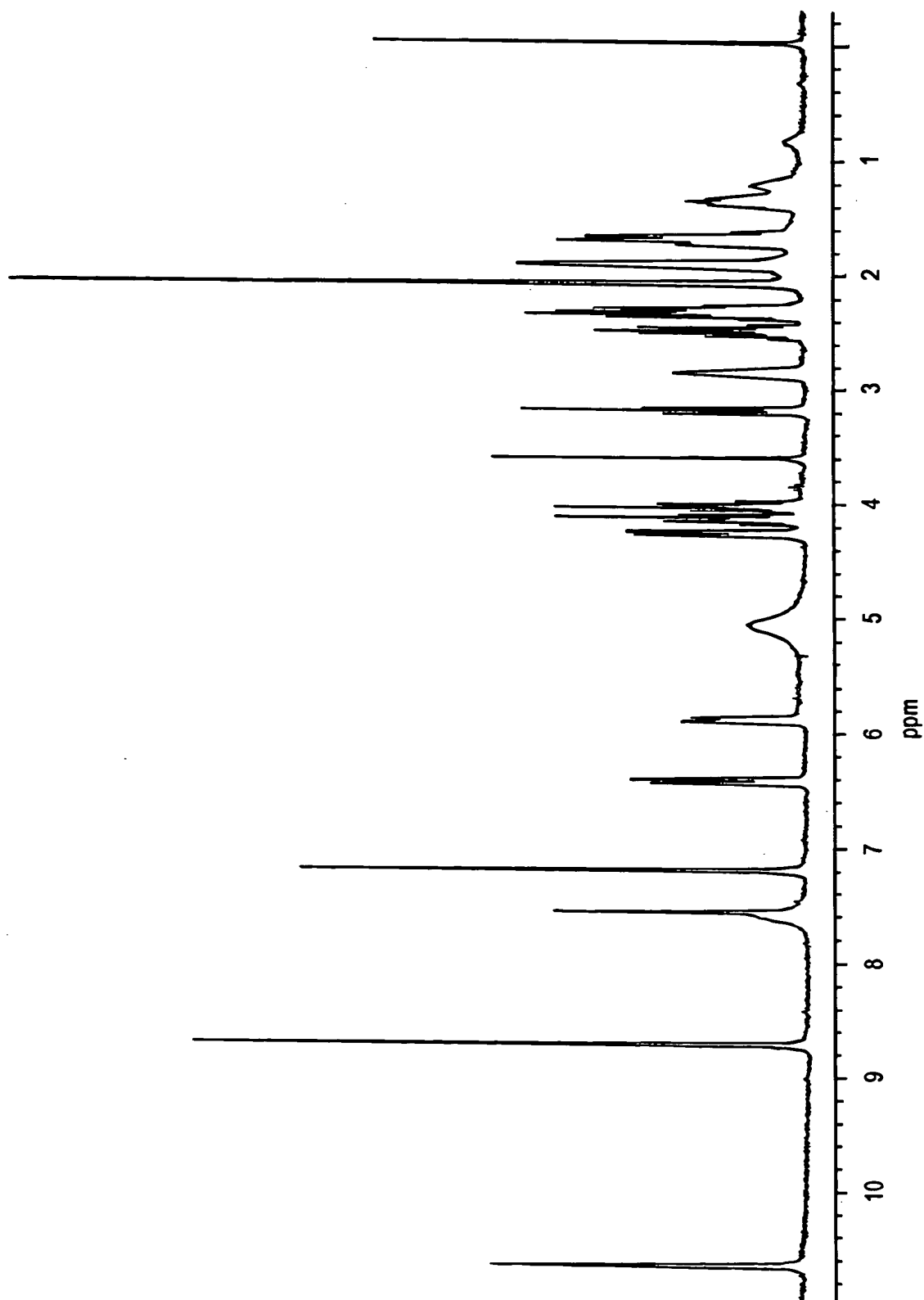
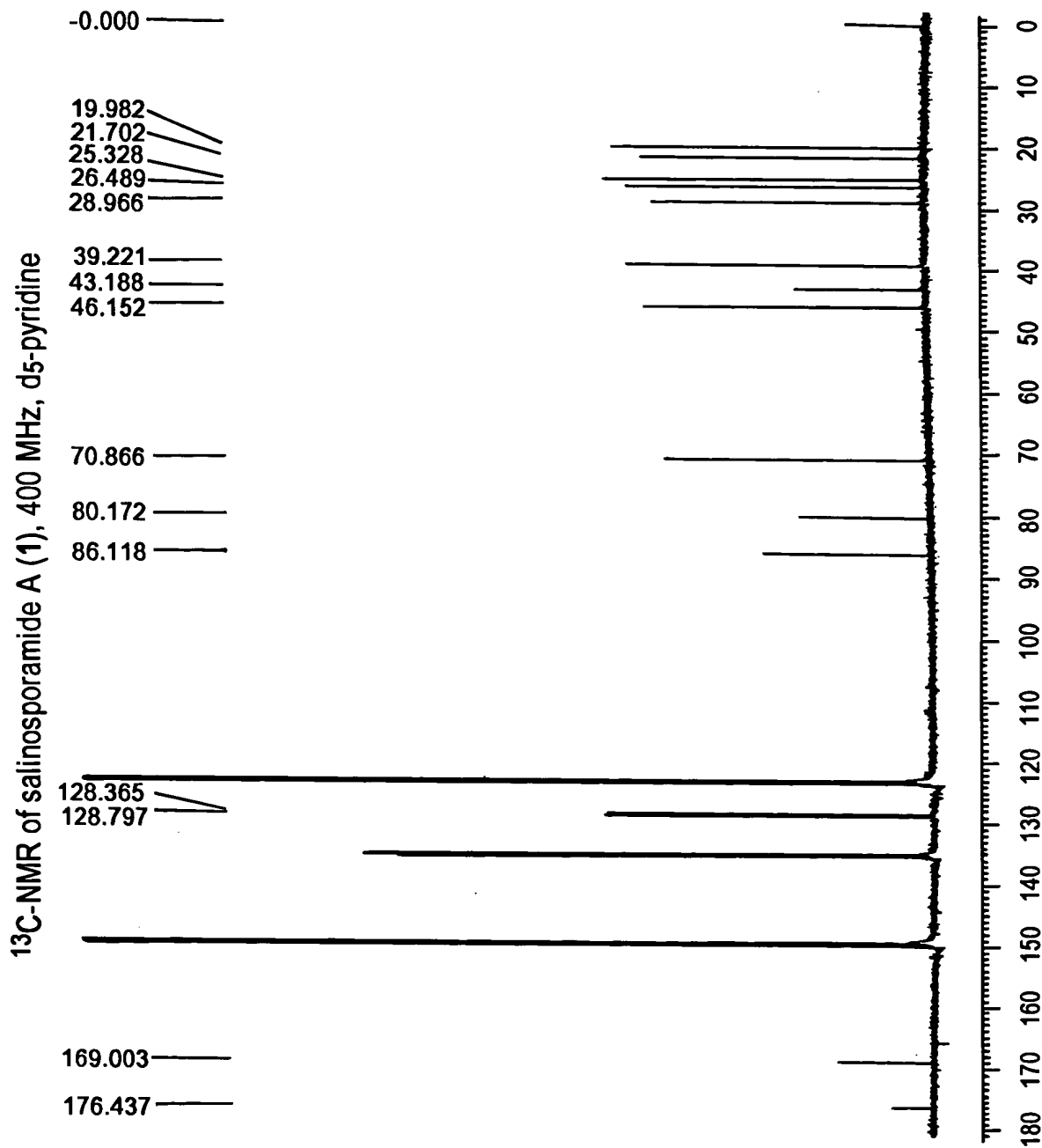


FIG. 6

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HMQC of salinosporamide A (1), 300 MHz, d₅-pyridine

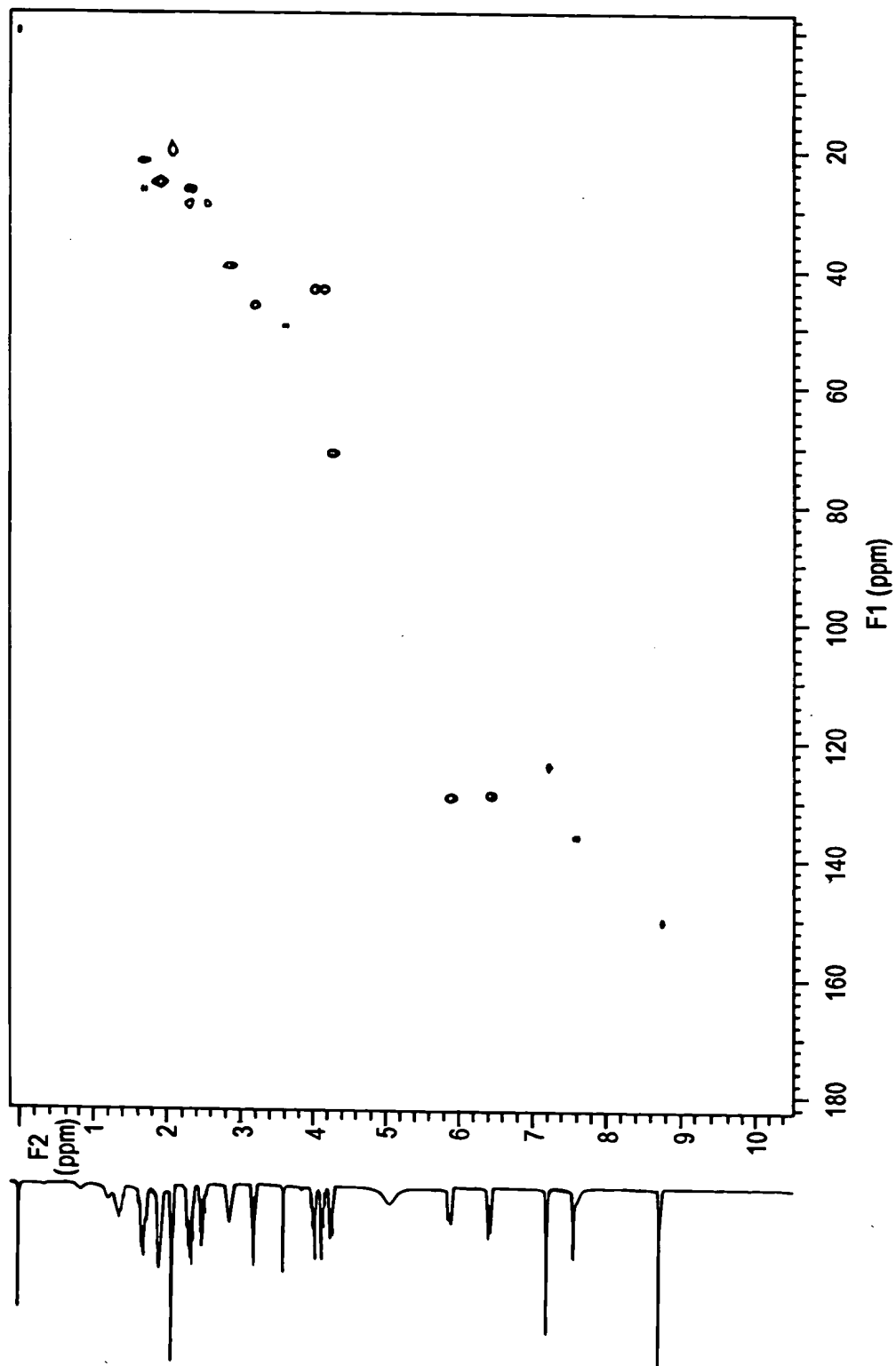


FIG. 8

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HMBC of salinosporamide A (1), 300 MHz, d₅-pyridine

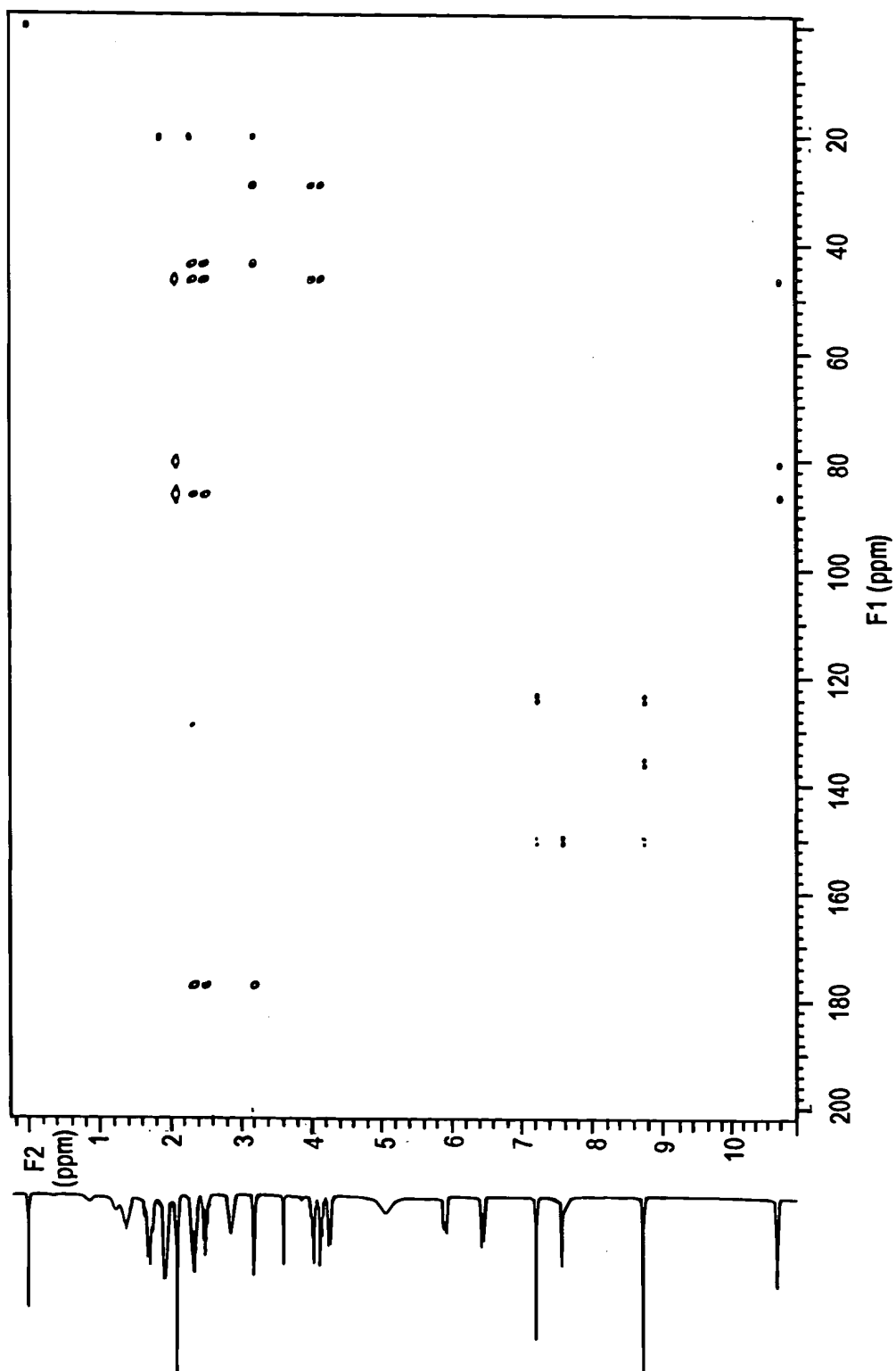


FIG. 9

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H,H-COSY of salinosporamide A (1), 300 MHz, d₅-pyridine

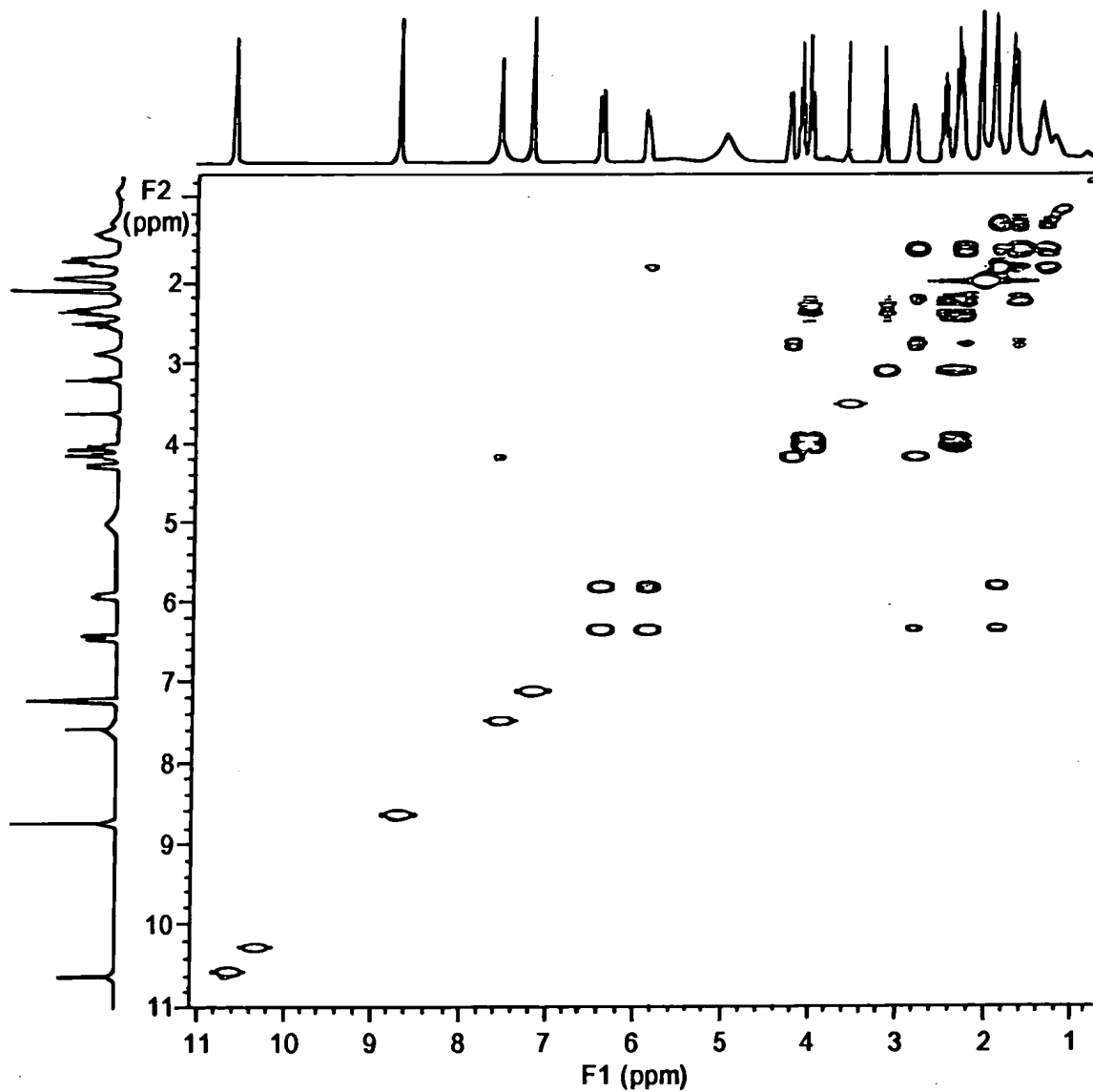


FIG. 10

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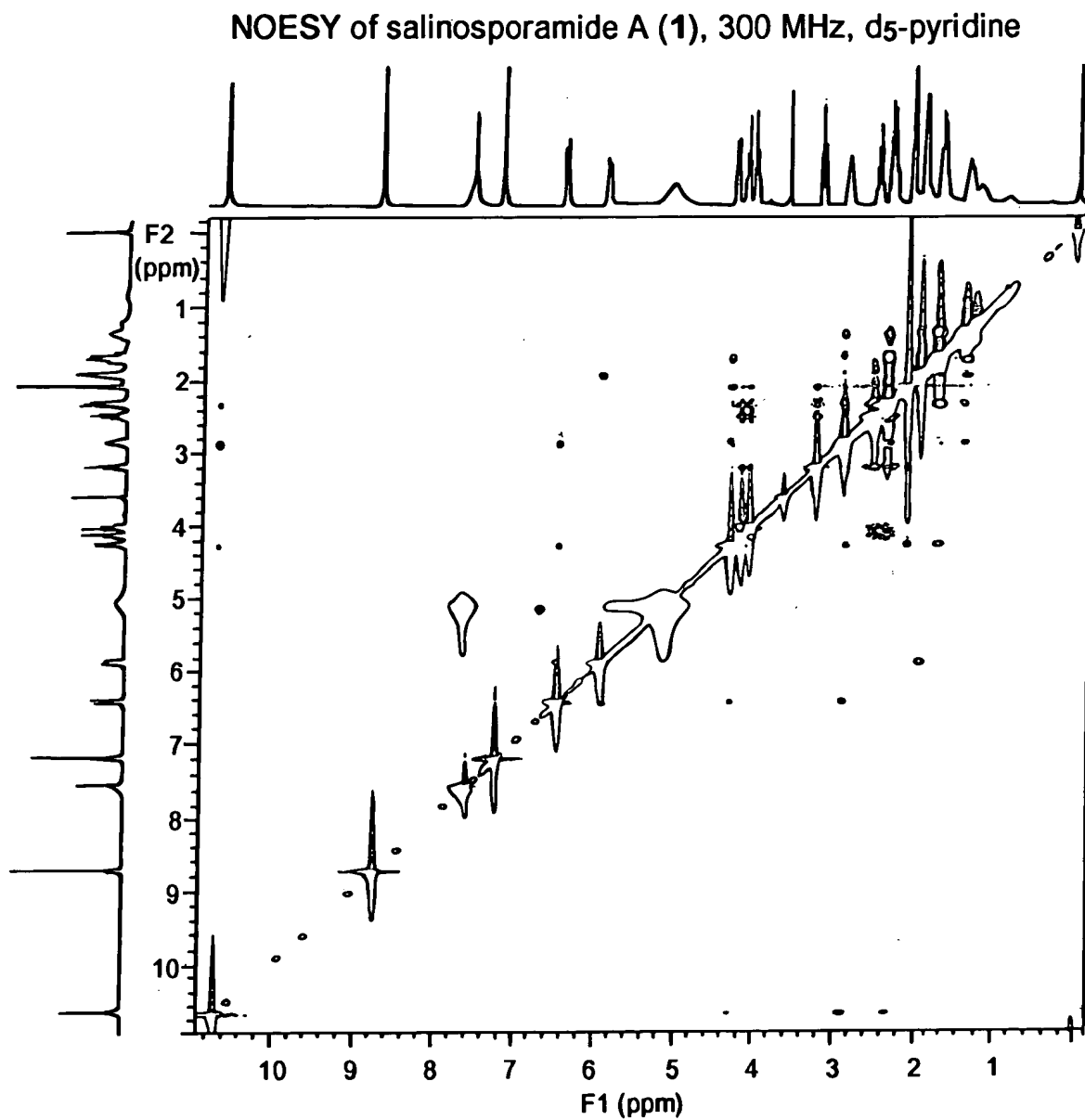


FIG. 11

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Mass spectrum (ESI-mode) of salinosporamide A (1)

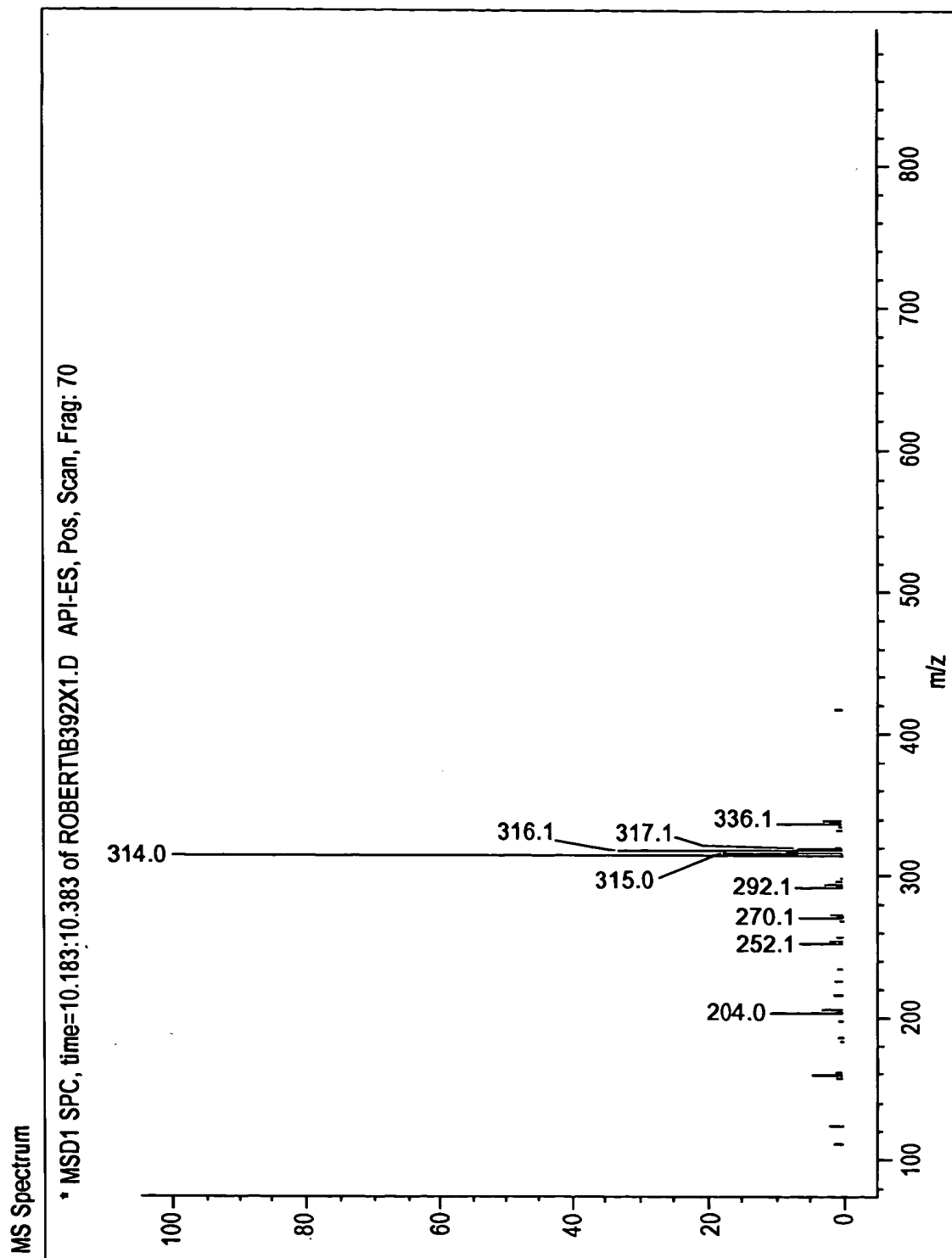


FIG. 12

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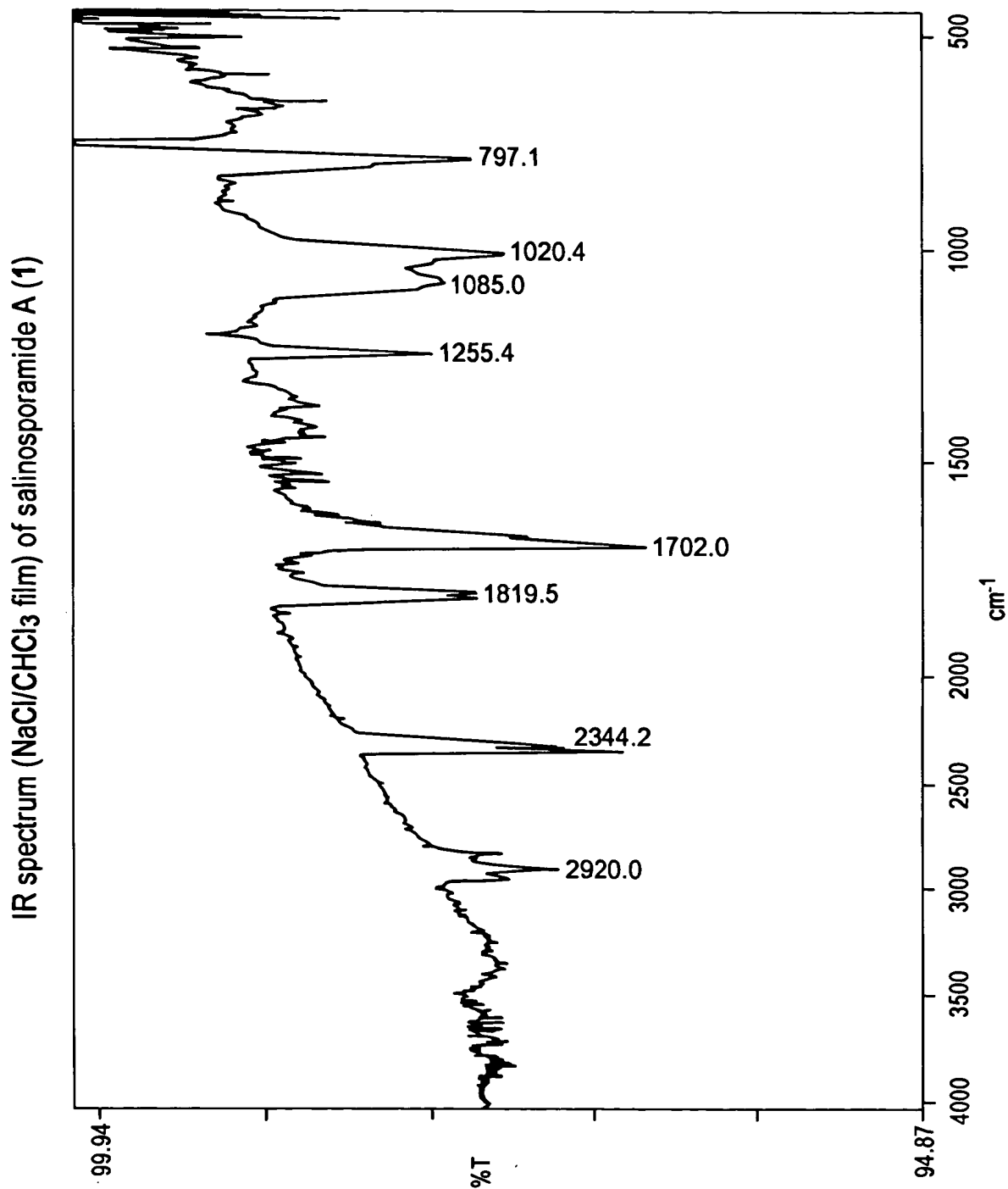


FIG. 13

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UV spectrum (LC-DAD, CH₃CN:water 2:1) of salinosporamide A (1)

DAD1, 11.674:11.807 (40.1 mAU, %Fs) Ref=2.927 & 12.007 of B392XA2.D

*DAD1, 11.674:11.807 (40.1 mAU, %Fs) Ref=2.927 & 12.007 of B392XA2.D

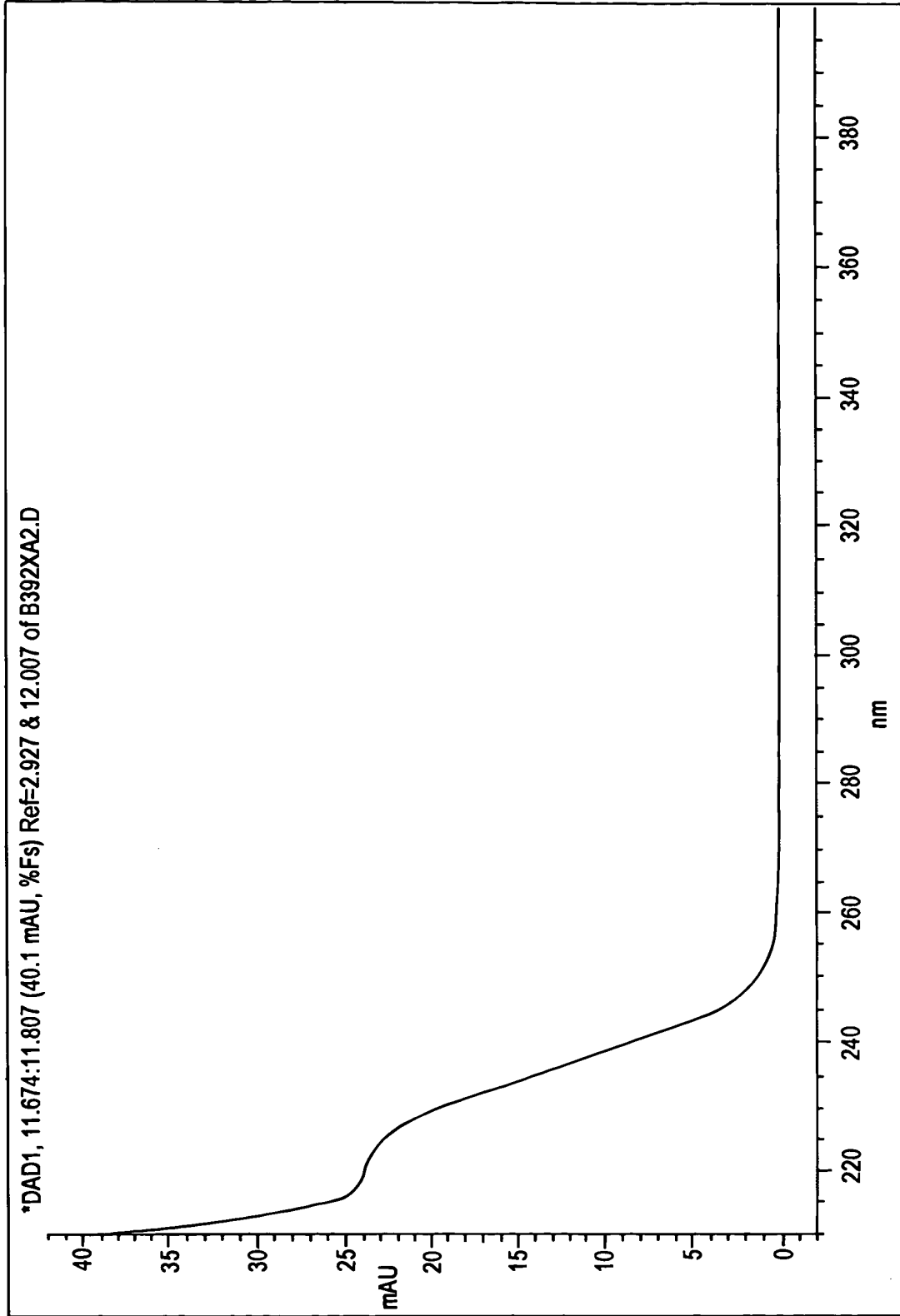


FIG. 14

Position 16S rDNA	Actinoplanes	Micromon- ospora	Dactylo- sporangium	Catellato- spora	Pillimelia	Catenulo- planes	Couchio- planes	Spirilli- planes	Verrucosi- spora	Marinospora
207	U	U	U	U	C	U	U	U	U	A
219	C	C	C	C	U	C	C	C	C	U
279	A	A	A	(AU)	A	A	A	A	A	G
366	A	(AG)	A	A	A	A	A	A	A	C
467	A	A	(GA)	A	G	(GA)	A	A	A	U
468	A	A	A	A	A	A	A	A	A	U
546	G	G	G	G	A	G	G	G	G	A
615	C	(GC)	C	(UC)	U	(CU)	C	C	U	U
1116	C	C	(CU)	C	C	C	C	C	C	U
1456	A	A	A	A	A	A	A	A	A	G

FIG. 15